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Unpicking the determinants of amide NH••O=C hydrogen bond strength with diphenylacetylene molecular balances

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Hydrogen bonding plays an essential part in dictating the properties of natural and synthetic materials. Secondary amides are well suited to cross–strand interactions through the display of both hydrogen bond donors and acceptors and are prevalent in polymers such as proteins, nylon, and Kevlar™. In attempting to measure hydrogen bond strength and to delineate the stereoelectronic components of the interaction, context frequently becomes vitally important. This makes molecular balances – systems in which direct comparison of two groups is possible – an appealing bottom up approach that allows the complexity of larger systems to be stripped away. We have previously reported a family of single molecule conformational switches that are responsive to diverse stimuli including Brønsted and Lewis acids, anions, and redox gradients. In this work we assess the ability of the scaffold, based on a 2,6-disubstituted diphenylacetylene, to measure accurately the difference in hydrogen bond strength between variously functionalised amides. In all of the examples investigated hydrogen bond strength closely correlate to measures of Brønstead acidity suggesting that the scaffold is well-suited as a platform for the accurate determination of bond strength in variously substituted systems.

Introduction

Hydrogen bonding is a critical determinant of structure, and thus function, in biological[1] and synthetic systems,[2] playing significant roles in properties such as solvation[3] and membrane permeability.[4] The nature of H–bonding has been debated in the literature since at least the 1930s.[5] IUPAC currently defines an H-bond broadly,[6] listing various forms of structural, spectroscopic, and computational evidence that may suggest that the interaction is present (Eq. 1).[7]

$$A - H + B \rightleftharpoons A - H \cdots B \qquad \text{(Eq. 1)}$$

However, recognizing the presence of an H–bond (HB) is of limited use in predicting how the relative strengths among multiple H–bond donors can affect conformational dynamism in natural and synthetic systems.[8,9] To answer this question a precise understanding of the factors underlying HB strength is required. Large–scale database searches, as well as numerous experimental and theoretical scales, have attempted to elucidate relative energies of hydrogen bonds and determine their effect on conformation.[10–12]

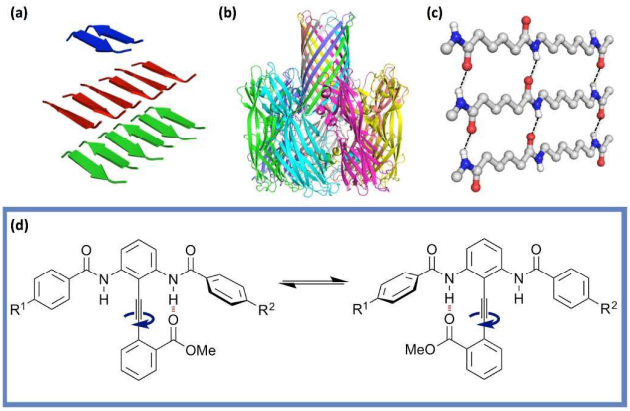

Fig. 1 Cross-strand hydrogen bonds between amides. Importance in medicine, technology and industry: (a) Aß plaques (pdb: 3ow9),[13] (b) A nanopore (pdb: 3m2l),[14] (c) Nylon 6,6 (ccdc: 1176016, H-bonds as dashed black lines);[15] (d) A molecular balance to measure H–bonding strength: two donors compete for a single acceptor. The equilibrium conformational ratio is determined by the relative H bond donor ability of the two benzamide NHs. Varying groups R^1 and R^2 allows probing of subtle steric and electronic effects.

Some of the earliest work in quantitative description of molecular properties was by Hammett, who described the aqueous ionization equilibria of substituted benzoic acids.[16] This work was expanded specifically for measuring H–bonding by Gurka and Taft, who developed the pK_{HB} scale of H-bond basicity using ^{19}F NMR studies of the complexation of bases with *p*–fluorophenol in carbon tetrachloride.[17] This H–bond basicity was found to be independent of aqueous pK_a, although the two scales are linearly correlated for a given functional group differing only in the electronic character of

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substituents.[18] An expanded scale was developed by Abraham and colleagues to describe 1:1 complexation of a variety of solutes in carbon tetrachloride.[19,20] Using this system, H–bond donors are described by a log K_A^H scale (alternatively described as α_2^H) and bases by a log K_B^H (or β_2^H) scale. These were subsequently expanded to include other stoichiometries, leading to $\Sigma\alpha_2^H$ and $\Sigma\beta_2^H$ scales.[21] Studies on hundreds of molecules indicate that these H–bond scales are likewise distinct from Brønsted acidity. More recently, theoretical descriptions of H–bonding have been developed using a variety of computational methods,[22–25] with varying degrees of success.

Of particular use would be a system in which two or more H–bond donors could be compared directly, giving internally consistent experimental evidence of the relative H–bond strengths. We recently reported the synthesis of molecular switches based on a diphenylacetylene (DPA), or tolan(e), scaffold[26] in which the relative HB strength of two amide NHs is the key determinant of conformation (Fig. 1d).[27–31] Experimental[32,33] and computational[34–40] methods are consistent with a small barrier for the relative rotation of two phenyl rings about an acetylene (≈ 0.6 – 0.8 kcal·mol^{-1} at room temperature), and single crystal X-ray diffraction indicate a separation of 3.0 – 3.2 Å between nitrogen and oxygen for the N-H••O=C HB arrangement of a 2,6–difunctionalised scaffold.

Whilst our previous work focussed on the use of external stimuli to moderate HB donor ability, and thus elicit large changes in conformational ratio, the system also has potential as a molecular balance for the direct measurement of relative HB strength. Through comparison of variously functionalised amides (Fig. 1d, R^1/R^2) there is scope to unravel structural and conformational determinants. Following Wilcox' seminal development of a molecular torsion balance,[41] Diederich,[42,43] Cockroft,[44,45] and others,[46] have shown that single molecule systems offer fundamental insights into various non–covalent interactions in molecular recognition events.

In order to test this hypothesis we set out to derive free energy differences from the conformational ratios of: (i) our previously published molecular switches; and (ii) expand the data set by synthesising and evaluating a library of novel compounds.

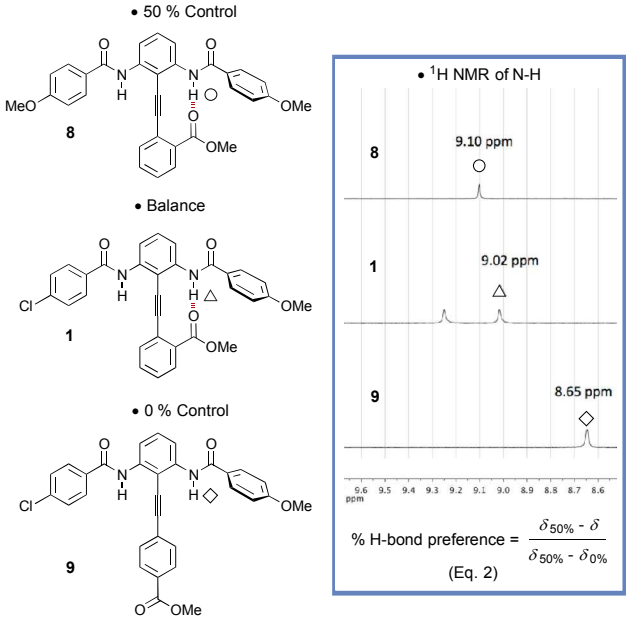
Results and discussion

A series of DPAs incorporating a variety of electron–withdrawing and electron–donating groups in positions R^1 and R^2 (balances **1** – **7**), along with 0 % and 50 % control compounds, were synthesised according to previously described methods (Table 1, columns 1–3).[27,28,†] Taking balance **1** (R^1 = Cl, R^2 = OMe) as an example, the solution phase conformational ratio is estimated by comparing the ^{1}H NMR amide NH resonances with those of two control compounds (Fig. 2). Control compound **8** juxtaposes the two *p*–methoxy–benzamides, and due to the symmetry of the system, the H–bond acceptor partitions equally between the two NH donors. Thus the amide resonance should estimate the chemical shift of the *p*–methoxybenzamide of **1** when the equilibrium ratio is 1:1 (50 % control). The second control compound **9** balances *p*–chloro– and *p*–methoxy–benzamide, similarly to **1** however the HB acceptor is now *para* to the alkyne linkage. Because the intramolecular HB has been removed, the *p*–methoxy amide ^{1}H resonance of **9** estimates the chemical shift of the same resonance in **1** if the H–bond equilibrium were biased completely toward the other side (0 % control).[47] The *p*–MeO–benzamide resonance of **1** (△) is downfield of the analogous resonance in **9** (◇), but upfield of that for **8** (○). This suggests that the *p*–MeO NH experiences more than 0 % but less than 50 % of the total H–bond interaction. Thus the position of the conformational equilibrium must be biased toward the *p*–Cl NH. Using the chemical shifts of these resonances the equilibrium ratio is estimated to be 1.43:1 toward the chloride substitution (Fig. 2, Eq. 2). The equilibrium ratio is related to the free energy difference of the two states by Eq. 3, in this case indicating a free energy difference for the two conformations of 0.21 kcal·mol^{-1}.

$$\Delta G = -RT\ ln(conformational\ ratio) \qquad \text{(Eq. 3)}$$

Free energy differences for compounds **2** – **7** were estimated using this method (Table 1, column 7).[†]

Fig. 2 Molecular balance **1**, with 50 % **8**, and 0 % **9**, control compounds. Inset: ^{1}H NMR conformational analysis. Unambiguous assignment of NH ^{1}H signals was performed by interpretation of nOe correlations with aryl *ortho*–hydrogens relative to the carbonyl group of the benzamide.[27]



In addition to the experimentally derived free energies, the molecules can also be described using various series of tabulated values of acidity or HB strength. For example, $\Delta\sigma_p$ gives the difference between the Hammett descriptor values of R^1 and R^2 (Fig. 1, and Eq. 4).

$$\Delta\sigma_p = \sigma^{R^2} - \sigma^{R^1} \qquad \text{(Eq. 4)}$$

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Organic & Biomolecular Chemistry

PAPER

Table 1 Calculated values for variously substituted diphenylacetylene molecular balances: Δ σ_p (pK_a), Δ σ_p (^{19}F), Δ log K_A^H; and Δ G values based on 50 % and 100 % controls.

Balance	R^1	R^2	Δ σ_p (pK_a)	Δ σ_p (^{19}F)	Δ log K_A^H	Δ G (50 %)	Δ G (100 %)
2	NMe_2	H	– 0.83	– 0.42	Not reported	– 0.17	– 0.16
3	OMe	H	– 0.27	– 0.40	– 0.11	– 0.03	– 0.10
4	H	H	0.00	0.00	0.00	0.00	0.00
5	H	Cl	+ 0.23	+ 0.21	+ 0.34	+ 0.17	+ 0.24
1	OMe	Cl	+ 0.50	+ 0.61	+ 0.45	+ 0.21	+ 0.22
6	H	NO_2	+ 0.78	+ 0.75	+ 1.05	+ 0.37	+ 0.45
7	OMe	NO_2	+ 1.05	+ 1.15	+ 1.16	+ 0.44	+ 0.59

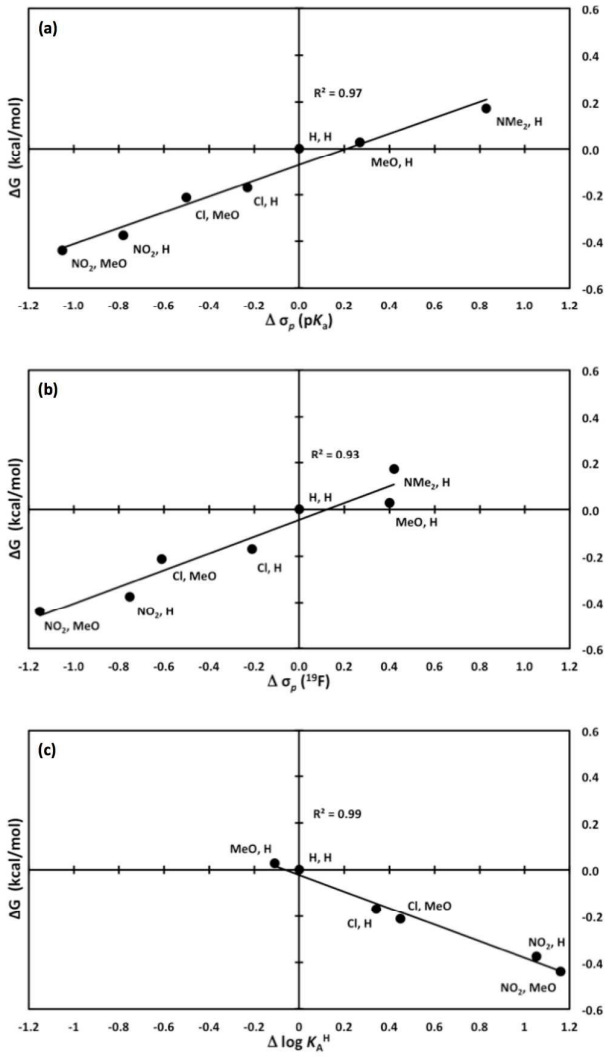

Fig. 3 Correlation between experimentally observed conformational free energy differences of variously substituted molecular balances with descriptors from: (a) aqueous Hammett ionization ratios; (b) ^{19}F NMR shifts; (c) 1:1 H–bond complex formation in carbon tetrachloride.

Table 1 gives the calculated $\Delta\sigma_p$ values using benzoic acid Hammett parameters, Hammett parameters calculated using ^{19}F chemical shifts of substituted 1–fluorobenzene,[48] and log K_A^H values from *p*–substituted phenols (columns **4 – 6**).[19] The experimentally derived equilibrium ratios are strongly correlated to the aqueous Hammett $\Delta\sigma_p$ values (r^2 = 0.97), ^{19}F–derived values measured in organic solvent (r^2 = 0.93), and for Δ log K_A^H measurements (r^2 = 0.99, Figs. 3a–c).[§] This indicates that in this system, HB strength among the homologous series of donors is highly correlated with metrics of Brønsted acidity.

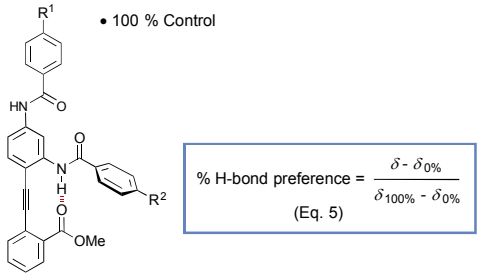

Fig. 4 Conformational analysis using a 100 % control compound.

Whilst the 0 % and 50 % control compounds appear to perform well, in order to exclude the possibility that the close correlation exhibited with Hammett parameters (Fig. 3) was due to fortuitous error cancellation, or unforeseen interference with our NMR assay, an alternative 100 % control molecule was designed and synthesized (Fig. 4). This molecule, which replaces the 50 % control, has similar electronic characteristics, but a markedly different conformation. The R^1 benzamide group is positioned *para* to the alkyne linkage, leaving only the benzamide R^2–functionalised donor available for HB formation. Using this control, the conformational ratio can then be determined by observing the amount a given NH resonance has shifted downfield of the 0 % control toward the 100 % control (Fig. 4, Eq. 5). The corresponding 100 % controls for compounds **2**, **3**, **5 – 7** were used to determine new free energy differences (Table 1, column 8. See supplementary information Chapter 4 for spectra and tabulated data of

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conformational ratios).† While the 50 % and 100 % data are similar for **1** and **2**, 100 % controls give slightly larger values for **3** and **5** – **7**. Despite this increase these new values produce a similar correlation with the reported descriptors (Fig. 5).

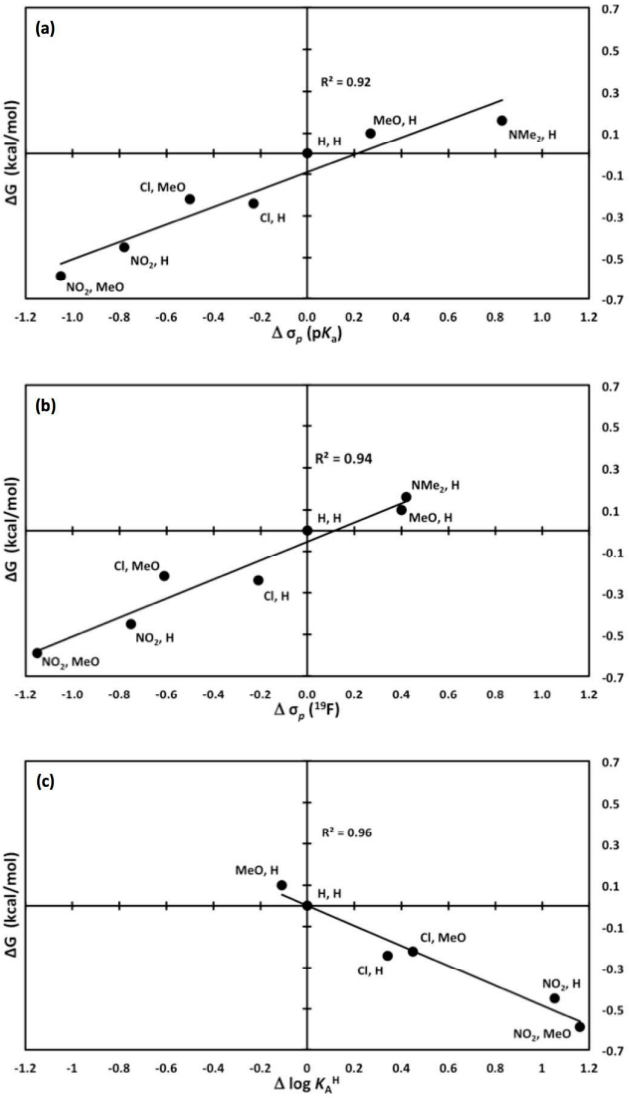

Fig. 5 Correlation between the observed conformational free energy difference of molecular balances using the 100 % control molecules with descriptors from: (a) aqueous Hammett ionization ratios; (b) ^{19}F NMR shifts; (c) 1:1 H–bond complex formation in carbon tetrachloride.

The level of correlation between free energies derived from conformational ratios and thermodynamic parameters is noteworthy given the steric clash inherent in H–bonding between the methyl benzoate and the benzamide ring (Fig. 1d). This rotation might be expected to reduce the π–overlap (σ_π) between the *para*–substituent and the amide carbonyl, and thus impact HB donor ability. However, the data fit very well to the unscaled, planar Hammett descriptors.

Previously reported dihedral angles between the carbonyl and aryl groups range between 36 ˚ and 52 ˚ in the solid–state.[27,28,30] In keeping with these values newly acquired diffraction data for **3** and **4** have dihedrals of 34 ˚ and 36 ˚ respectively (Fig. 6). Balance **3**, in which *p*–H competes with *p*-OMe, is a particularly interesting case in that the solid–state conformation adopted is opposite that of the solution–phase.

Table 2 Inductive and mesomeric components of Hammett parameters for *para*–substituted benzoic acids.[48]

	para–Substituent R				
	H	**Cl**	**NO_2**	**NMe_2**	**OMe**
σ_I	0.00	+ 0.43	+ 0.64	+ 0.17	+ 0.30
σ_π	0.00	– 0.16	+ 0.16	– 0.56	– 0.43

A plausible hypothesis is that attenuation of the mesomeric contribution (σ_π) for *p*–OMe due to sterics leads to a superior H–bond donor relative to *p*–H through the operation of inductive effects (σ_I), albeit by a small margin. This is consistent with the comparable magnitudes, and opposite in sign, of the mesomeric and inductive components for *p*–OMe (Table 2).[48]

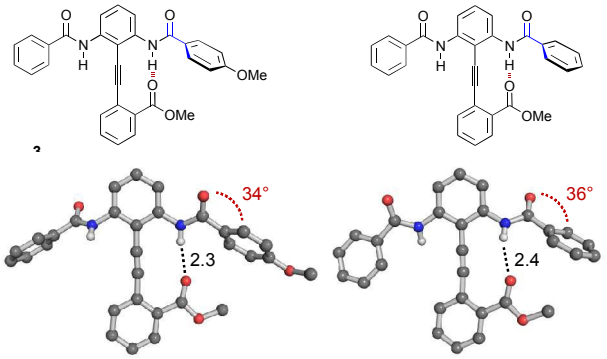

Fig. 6 The solid–state structures of balances **3** and **4** (CCDC 871789 and 871790).† The measured carbonyl–aryl dihedral angles are illustrated in blue on the structural formulae and the values are shown in red next to the diffraction images. The black dashed lines and black labels give H–bond lengths in Å.

To gain insights into the contrasting behaviour between solid– and solution–state data we sought a method to determine the effect of dihedral angle on H–bond strength in the solution phase. The ionization energies of *para*–substituted benzoic acids were determined using the high–level composite CBS–QB3 quantum chemical method.[49,†] The deprotonation energies for *p*–substituents H, Cl, NO_2, NMe_2, and OMe were calculated as a function of the dihedral angle between the carboxyl group and the aryl ring (Fig. 7, Table 3). The results do not appear to fit a simple function, but instead show very little variation in deprotonation energy until 40˚ is reached, at which point the energy difference becomes more pronounced. For example, the electron donating dimethylamino substituent of **2** experiences a maximal energy difference of – 3.23 kcal·mol^{-1} at 70˚ between the neutral and deprotonated forms. The methoxy substituted molecule has a + 0.52 kcal·mol^{-1} energy difference relative to benzoic acid at 90˚, but intriguingly only a + 0.08 kcal·mol^{-1} difference at the maximum crystallographically observed 50˚ dihedral.†

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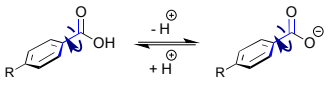

Fig. 7 Calculation of deprotonation energy for *para*–substituted benzoic acids as a function of dihedral angle (indicated in blue). See also Table 3.

Table 3 Calculated deprotonation energies for *para*–substituted benzoic acids as a function of dihedral angle relative to the planar conformation.

Dihedral	ΔE / kcal•mol^{-1}, *para*–substituent R				
angle	H	Cl	NO_2	NMe_2	OMe
0	0.00	0.00	0.00	0.00	0.00
10	– 0.01	– 0.01	+ 0.02	– 0.34	– 0.01
20	– 0.03	– 0.03	– 0.09	– 0.42	– 0.03
30	– 0.08	– 0.09	– 0.24	– 0.64	– 0.09
40	– 0.18	– 0.23	– 0.47	– 0.96	– 0.22
50	– 0.97	– 1.00	– 1.36	– 1.95	– 1.05
60	– 0.59	– 0.58	– 1.11	– 1.83	– 0.84
70	– 0.96	– 0.98	– 1.44	– 3.23	– 1.25
80	– 1.19	– 1.22	– 1.70	– 2.64	– 1.64
90	– 1.25	– 1.26	– 1.66	– 1.19	– 1.77

The magnitude of values from the calculations suggests that all dihedral angles are thermally accessible at room temperature. Moreover the effect of rotating the benzamide involved in the hydrogen bond is relatively constant, up to approximately 40 ˚, despite a range of substituents. Thus this small effect exists essentially as a constant in the linear free energy relationships and is not large enough to significantly bias the equilibrium conformational ratios of these molecules (Table 1, Figs. 3 and 5). This hypothesis is supported by previously reported studies on the ^{13}C NMR substituent chemical shifts (SCS) of 4–substituted–2,6–dimethyl benzamides.[50] Solid–state analysis of these dimethyl benzamides shows that the aryl ring is rotated out of the amide plane by ≈ 56 ˚ and is accompanied by only partial loss of conjugation.†

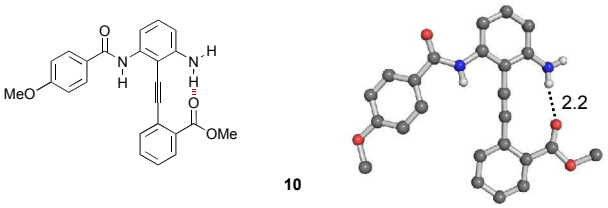

Fig. 8 The solid–state structure of mono–substituted benzamide **10** (CCDC 1565855).† The black dashed lines and black labels give H–bond lengths in Å.

The computational analysis (Table 3) is consistent with the goodness of fit between our experimentally determined conformational ratios in the solution–phase and unscaled Hammett data. Returning to the reversal of H–bond donor preference in the solid–state for balance **3**, in the absence of aryl–carbonyl free rotation, attenuation of mesomeric donation (σ_π) likely leads to a slight preference in favour of inductive withdrawal (σ_I) and thus the *p*–H substituted benzamide becomes an inferior H–bond donor.[51,52] Further evidence for the importance of sterics in H–bond preference comes from solid–state data obtained for mono–substituted benzamide **10**, in which hydrogen bonding to the aniline is preferred over the potentially superior benzamide donor (Fig. 8).

Conclusions

Computation, solid–, and solution–phase conformational analysis demonstrate that a simple molecular balance based on a 2,6–disubstituted DPA allows direct comparison of HB strength in aryl amides. An operationally quick and straightforward ^1H NMR assay in organic solvent, using one of two sets of control compounds, provides data that correlate closely with tabulated parameters such as Hammett substituent constants measured in physiologically relevant aqueous media. An interesting insight gained from *ab initio* calculations of *p*–substituted benzoic acid acidity was the dependence of π–overlap on dihedral angle. For various inductive and mesomeric groups the effect was small (< 1 kcal·mol^{-1}) at angles of less than 40 ˚. The study also sounds a cautionary note when inferring solution–phase conformational preferences of H–bond systems from their solid–state data. Whilst crystal–packing forces are frequently cited as important determinants of solid–phase conformation the underlying stereoelectronics are often difficult to unravel. Future work will explore the extension of the balance to aqueous solvents with heterocyclic, aliphatic and amino acid derived amides.

Experimental details

The synthesis and conformational behaviour, relative to 0 % and 50 % controls, of **2**, **4**,‡ **5** and **6** have previously been reported.[27,28] For full experimental details describing: (i) the synthesis of precursors; (ii) the synthesis of 0 % and 50 % control molecules for balances **3** and **7**; (iii) the synthesis of 100 % control molecules for balances **1**–**3** and **5**–**7**; (iv) conformational analysis; and (v) computation, please refer to the ESI.†

Synthetic procedures

Methyl 2–((2–(4–chlorobenzamido)–6–(4–methoxybenza–mido)phenyl)ethynyl)benzoate (1). 4–Dimethylaminopyridi–ne (0.1 mg) was added to a solution of methyl 2–((2–amino–6–(4–methoxybenzamido)phenyl)ethynyl)benzoate **10**† (100 mg, 0.25 mmol) in dichloromethane (0.05 M). Pyridine (23 μL, 0.28 mmol, 1.1 eq.) was added and the solution was stirred for 10 mins. Freshly prepared 4–chlorobenzoyl chloride **S8**† (71 μL, 0.55 mmol, 2.2 eq.) was added dropwise over 1 min and the solution stirred for 18 h. The reaction mixture was poured into 2 *N* hydrochloric acid (10 mL), extracted with dichloromethane (3 x 10 mL), and washed with 2 *N* sodium hydroxide (10 mL). The organic layers were dried over sodium sulfate, and concentrated *in vacuo*. The residue was purified by recrystallization from refluxing acetonitrile/water to give the *title compound* **1** as a white solid (90 mg, 0.27 mmol, 66 %):

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δ_H (400 MHz, $CDCl_3$) 9.25 (s, 1H), 9.02 (s, 1H), 8.36 (d, *J* 8.3, 1H), 8.32 (d, *J* 8.3, 1H), 8.10 (d, *J* 7.8, 1H), 7.96 (d, *J* 8.8, 2H), 7.93 (d, *J* 8.5, 2H), 7.60 (d, *J* 4.0, 2H), 7.49 (m, 2H), 7.44 (d, *J* 8.4, 2H), 6.98 (d, *J* 8.7, 2H), 3.89 (s, 3H), 3.52 (s, 3H); δ_C (126 MHz, $CDCl_3$) 165.6, 165.5, 165.4, 162.7, 140.3, 140.1, 138.0, 133.9, 133.1, 132.6, 131.2, 131.1, 130.3, 129.5, 129.4, 129.0, 128.7, 127.4, 123.1, 115.2, 115.2, 113.9, 102.88, 102.4, 86.0, 77.4, 77.2, 76.9, 55.6, 52.1; HRMS (ESI): found 539.1359; $C_{31}H_{23}ClN_2O_5$ $[M + H]^+$ requires: 539.1368.

Methyl 2–((2–benzamido–6–(4–methoxybenzamido)ph–enyl)ethynyl)benzoate (3).‡ Based on the procedure for **1**, methyl 2–((2–amino–6–(4–methoxybenzamido) phenyl)ethyn–yl)benzoate **10**† and benzoyl chloride **S9**† gave the *title compound* **3** as a white solid (95 mg, 0.18 mmol, 70 %): δ_H (500 MHz, $CDCl_3$) 9.21 (s, 1H), 9.06 (s, 1H), 8.32 (dd, *J* 4.9, 8.3, 2H), 8.05 (d, *J* 7.9, 1H), 7.95 (dd, *J* 8.6, 9.8, 4H), 7.54 (m, 3H), 7.44 (dd, *J* 7.4, 15.1, 4H), 6.94 (d, *J* 7.9, 2H), 3.86 (d, *J* 0.9, 3H), 3.44 (d, *J* 1.0, 3H); δ_C (126 MHz, $CDCl_3$) 166.4, 165.7, 165.3, 162.7, 140.4, 140.3, 135.4, 133.1, 132.5, 132.3, 131.9, 131.1, 131.0, 130.3, 129.6, 128.9, 128.6, 127.8, 127.5, 123.2, 115.2, 115.1, 102.9, 102.4, 86.1, 55.5, 52.2; HRMS (ESI): found 527.1568; $C_{31}H_{24}N_2O_5$ $[M + Na]^+$ requires 527.1577.

Methyl 2–((2–(4–methoxybenzamido)–6–(4–nitroben–zamido)phenyl)ethynyl)benzoate (7). Based on the procedure for **1**, methyl 2–((2–amino–6–(4–methoxybenzamido)phenyl)–ethynyl)benzoate **10**† and 4–nitrobenzoyl chloride gave the *title compound* **7** as a pale yellow solid (64 mg, 0.12 mmol, 47 %) after precipitation from refluxing acetonitrile/water: δ_H (400 MHz, $CDCl_3$) 9.44 (s, 1H), 8.95 (s, 1H), 8.40 (d, *J* 8.4, 1H), 8.32 (dd, *J* 4.9, 11.7, 3H), 8.17 (m, 2H), 8.10 (d, *J* 7.8, 1H), 7.96 (m, 2H), 7.60 (m, 2H), 7.51 (m, 2H), 7.00 (d, *J* 8.9, 2H), 3.90 (s, 3H), 3.49 (s, 3H); δ_c (126 MHz, $CDCl_3$) 165.6, 165.3, 164.9, 162.8, 149.7, 141.2, 140.3, 139.9, 133.0, 132.8, 131.3, 131.2, 130.4, 129.4, 129.2, 127.3, 123.6, 123.0, 115.6, 115.4, 114.1, 103.0, 102.6, 85.8, 55.6, 52.2; HRMS (ESI): found 550.1605; $C_{31}H_{23}N_3O_7$ $[M + H]^+$ requires 550.1609.

Methyl 2–((2,6–bis(4–methoxybenzamido)phenyl)ethyn–yl)benzoate (8). Based on the procedure for **1**, methyl 2–((2–amino–6–(4–methoxybenzamido)phenyl)ethynyl)benzoate **10**† and 4–methoxybenzoyl chloride **S3**† gave the *title compound* **8** as a white solid (108 mg, 0.20 mmol, 81 %) following recrystallization from refluxing acetonitrile/water: δ_H (500 MHz, $CDCl_3$) 9.10 (s, 2H), 8.33 (d, *J* 8.3, 2H), 8.11 (d, *J* 7.7, 1H), 7.97 (d, *J* 8.8, 4H), 7.65 – 7.56 (m, 2H), 7.49 (t, *J* 5.4, 2H), 6.97 (d, *J* 8.8, 4H), 3.89 (s, 6H), 3.55 (s, 3H); δ_C (101 MHz, $CDCl_3$) 165.7, 165.4, 162.6, 140.3, 133.1, 132.6, 131.1, 131.0, 130.3, 129.6, 128.9, 127.5, 123.2, 114.9, 113.8, 102.8, 102.2, 86.1, 55.6, 52.2; HRMS (ESI): found 535.1857; $C_{32}H_{26}N_2O_6$ $[M + H]^+$ requires 535.1864.

Methyl 4–((2–(4–chlorobenzamido)–6–(4–methoxybenza–mido)phenyl)ethynyl)benzoate (9). Based on the procedure for **1**, methyl 4–((2–amino–6–(4–methoxybenzamido)phenyl)–ethynyl)benzoate **S13**† and 4–chlorobenzoyl chloride **S8**† gave the *title compound* **9** as a yellow solid (70 mg, 0.13 mmol, 52 %): δ_H (500 MHz, $CDCl_3$) 8.65 (s, 2H), 8.38 (d, *J* 8.3, 1H), 8.32 (d, *J* 8.3, 1H), 8.13 (d, *J* 8.0, 2H), 7.91 (dd, *J* 8.5, 11.5, 4H), 7.60 (d, *J* 8.0, 2H), 7.50 (d, *J* 8.6, 3H), 7.00 (d, *J* 8.4, 2H), 3.98 (s, 3H), 3.91 (s, 3H); δ_C (101 MHz, $CDCl_3$) 166.2, 164.8, 164.2, 163.0, 139.8, 139.2, 138.8, 133.2, 131.5, 131.2, 131.0, 130.2, 129.4, 129.0, 128.5, 126.9, 125.9, 115.4, 115.1, 113.3, 103.4, 101.8, 83.1, 55.7, 52.7;HRMS (ESI): found 539.1372; $C_{31}H_{23}ClN_2O_5$ $[M + H]^+$ requires 539.1368.

Methyl 2–((2–amino–6–(4–methoxybenzamido)phenyl)–ethynyl)benzoate (10).‡ *N*–(3–amino–2–iodophenyl)–4–met–hoxybenzamide **S5**† (541 mg, 1.47 mmol) was added to a stirred solution of methyl 2–ethynylbenzoate **S6**† (260 mg, 1.62 mmol, 1.1 eq.) in anhydrous triethylamine (0.15 M) and *N*,*N*–dimethylformamide (0.15 M). The solution was degassed by sparging with nitrogen for 10 mins after which copper(I) iodide (25 mg, 0.13 mmol, 10 mol%) and palladium(II) chloride bis(triphenylphosphine) (63 mg, 0.09 mmol, 6 mol%) were each added as a single portion. Following 2 h at 60 ˚C the solution was allowed to cool, filtered over Celite™ and concentrated *in vacuo*. The residue was purified by flash column chromatography on silica gel (9:1 $CHCl_3$/EtOAc) to give the *title compound* **10** (472 mg, 1.18 mmol, 80 %) as a yellow solid: δ_H (500 MHz, $CDCl_3$) 8.96 (s, 1H), 8.06 (d, *J* 7.9, 1H), 7.95 (d, *J* 8.7, 2H), 7.92 (d, *J* 8.3, 1H), 7.58 (d, *J* 7.6, 1H), 7.53 (t, *J* 7.5, 1H), 7.39 (t, *J* 7.6, 1H), 7.19 (t, *J* 8.2, 1H), 6.97 (d, *J* 8.7, 2H), 6.49 (d, *J* 8.1, 1H), 4.86 (s, 2H), 3.88 (s, 3H), 3.78 (s, 3H); δ_C (126 MHz, $CDCl_3$) 165.9, 165.4, 162.5, 149.7, 140.2, 133.1, 132.4, 131.2, 131.0, 130.0, 129.4, 128.0, 127.9, 124.1, 113.9, 109.5, 108.6, 101.4, 97.5, 87.9, 55.6, 52.4; HRMS (ESI): found 401.1488; $C_{24}H_{20}N_2O_4$ $[M + H]^+$ requires 401.1496.

Conformational analysis

The position of the conformational equilibrium exhibited by balances **1** – **7** was examined by comparison of the position of the ^{1}H NMR signal corresponding to the R^1 or R^2 benzamide NH with its position in the corresponding 0 % and 50 % control molecules (Eq. 2), or by comparison with the 0 % and 100 % control molecules (Eq. 5). See the ESI† for spectra.

CBS–QB3 quantum chemical method

The protonated and deprotonated form of each carboxylic acid were built in GaussView[53] and analysed using a modredundant optimization with the carbonyl dihedral angle fixed to the specified value relative to the plane of the phenyl ring. Calculations were performed in the gas phase using the composite CBS–QB3 method[49] using the default parameters in Gaussian09.[54,†]

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Notes and references

‡ The X–ray data for **3**, **4**, and **10** has been deposited in the Cambridge Crystallographic Data Centre (CCDC 871789, 871790 & 1565855).

§ The value for NMe_2 was not reported and thus the fit is through one fewer point.[19]

§§ Chloro was not reported.

1 G. A. Jeffrey and W. Saenger, *Hydrogen Bonding in Biological Structures*, Springer Science & Business Media, 2012.
2 P. M. Pihko, *Hydrogen Bonding in Organic Synthesis*, Wiley-VCH Verlag GmbH & Co. KGaA, 2009.
3 H.-P. Cheng, *J. Phys. Chem. A*, 1998, **102**, 6201–6204.
4 J. M. Boggs, *BBA Biomembranes*, 1987, **906**, 353–404.
5 L. Pauling, *The Nature of the Chemical Bond and the Structure of Molecules and Crystals: An Introduction to Modern Structural Chemistry*, Cornell University Press, 1st edition., 1939.
6 The hydrogen bond is an attractive interaction between a hydrogen atom from a molecule or a molecular fragment X–H in which X is more electronegative than H, and an atom or a group of atoms in the same or a different molecule, in which there is evidence of bond formation.
7 E. Arunan, G. R. Desiraju, R. A. Klein, J. Sadlej, S. Scheiner, I. Alkorta, D. C. Clary, R. H. Crabtree, J. J. Dannenberg, P. Hobza, H. G. Kjaergaard, A. C. Legon, B. Mennucci and D. J. Nesbitt, *Pure Appl. Chem.*, 2011, **83**, 1637–1641.
8 P. C. Knipe, S. Thompson and A. D. Hamilton, *Chem. Sci.*, 2015, **6**, 1630–1639.
9 F. Giordanetto, C. Tyrchan and J. Ulander, *ACS Med. Chem. Lett.*, 2017, **8**, 139–142.
10 B. Kuhn, P. Mohr and M. Stahl, *J. Med. Chem.*, 2010, **53**, 2601–2611.
11 C. Bissantz, B. Kuhn and M. Stahl, *J. Med. Chem.*, 2010, **53**, 5061–5084.
12 P. Gilli, L. Pretto, V. Bertolasi and G. Gilli, *Acc. Chem. Res.*, 2009, **42**, 33–44.
13 J.-P. Colletier, A. Laganowsky, M. Landau, M. Zhao, A. B. Soriaga, L. Goldschmidt, D. Flot, D. Cascio, M. R. Sawaya and D. Eisenberg, *Proc. Natl. Acad. Sci. USA*, 2011, **108**, 16938–16943.
14 A. Banerjee, E. Mikhailova, S. Cheley, L.-Q. Gu, M. Montoya, Y. Nagaoka, E. Gouaux and H. Bayley, *Proc. Natl. Acad. Sci. USA*, 2010, **107**, 8165–8170.
15 C. W. Bunn and E. V. Garner, *Proc. R. Soc. A.*, 1947, **189**, 39–68.
16 L. P. Hammett, *J. Am. Chem. Soc.*, 1937, **59**, 96–103.
17 D. Gurka and R. W. Taft, *J. Am. Chem. Soc.*, 1969, **91**, 4794–4801.
18 R. W. Taft, D. Gurka, L. Joris, P. von R. Schleyer and J. W. Rakshys, *J. Am. Chem. Soc.*, 1969, **91**, 4801–4808.
19 M. H. Abraham, P. L. Grellier, D. V. Prior, P. P. Duce, J. J. Morris and P. J. Taylor, *J. Chem. Soc., Perkin Trans. 2*, 1989, 699–711.
20 M. H. Abraham, P. L. Grellier, D. V. Prior, J. J. Morris and P. J. Taylor, *J. Chem. Soc., Perkin Trans. 2*, 1990, 521–529.
21 M. H. Abraham, *Chem. Soc. Rev.*, 1993, **22**, 73–83.
22 J. A. Platts, *Phys. Chem. Chem. Phys.*, 2000, **2**, 973–980.
23 J. Ireta, J. Neugebauer and M. Scheffler, *J. Phys. Chem. A*, 2004, **108**, 5692–5698.
24 R. S. Paton and J. M. Goodman, *J. Chem. Inf. Model.*, 2009, **49**, 944–955.
25 K. Wendler, J. Thar, S. Zahn and B. Kirchner, *J. Phys. Chem. A*, 2010, **114**, 9529–9536.
26 S. Toyota, *Chem. Rev.*, 2010, **110**, 5398–5424.
27 I. M. Jones and A. D. Hamilton, *Org. Lett.*, 2010, **12**, 3651–3653.
28 I. M. Jones, H. Lingard and A. D. Hamilton, *Angew. Chem. Int. Ed.*, 2011, **50**, 12569–12571.
29 I. M. Jones, P. C. Knipe, T. Michaelos, S. Thompson and A. D. Hamilton, *Molecules*, 2014, **19**, 11316–11332.
30 P. C. Knipe, H. Lingard, I. M. Jones, S. Thompson and A. D. Hamilton, *Org. Biomol. Chem.*, 2014, **12**, 7937–7941.
31 P. C. Knipe, I. M. Jones, S. Thompson and A. D. Hamilton, *Org. Biomol. Chem.*, 2014, **12**, 9384–9388.
32 K. Okuyama, T. Hasegawa, M. Ito and N. Mikami, *J. Phys. Chem.*, 1984, **88**, 1711–1716.
33 K. Inoue, H. Takeuchi and S. Konaka, *J. Phys. Chem. A*, 2001, **105**, 6711–6716.
34 A. Liberles and B. Matlosz, *J. Org. Chem.*, 1971, **36**, 2710–2713.
35 A. Mavridis and I. Moustakali-Mavridis, *Acta. Cryst.*, 1977, **B33**, 3612–3615.
36 S. Saebø, J. Almlöf, J. E. Boggs and J. G. Stark, *Comp. Theor. Chem.*, 1989, **200**, 361–373.
37 A. Shimojima and H. Takahashi, *J. Phys. Chem.*, 1993, **97**, 9103–9112.
38 J. M. Seminario, A. G. Zacarias and J. M. Tour, *J. Am. Chem. Soc.*, 1998, **120**, 3970–3974.
39 Y. Li, J. Zhao, X. Yin and G. Yin, *ChemPhysChem*, 2006, **7**, 2593–2600.
40 Y. Li, J. Zhao, X. Yin, H. Liu and G. Yin, *Phys. Chem. Chem. Phys.*, 2007, **9**, 1186–1193.
41 S. Paliwal, S. Geib and C. S. Wilcox, *J. Am. Chem. Soc.*, 1994, **116**, 4497–4498.
42 F. R. Fischer, W. B. Schweizer and F. Diederich, *Angew. Chem. Int. Ed.*, 2007, **46**, 8270–8273.
43 F. R. Fischer, P. A. Wood, F. H. Allen and F. Diederich, *Proc. Natl. Acad. Sci. USA*, 2008, **105**, 17290–17294.
44 K. B. Muchowska, C. Adam, I. K. Mati and S. L. Cockroft, *J. Am. Chem. Soc.*, 2013, **135**, 9976–9979.
45 L. Yang, C. Adam, G. S. Nichol and S. L. Cockroft, *Nat. Chem.*, 2013, **5**, 1006–1010.
46 I. K. Mati and S. L. Cockroft, *Chem. Soc. Rev.*, 2010, **39**, 4195–4205.
47 Our previous studies discount intermolecular H-bonding since NH shifts are concentration invariant: *Org. Lett.*, 2010, **12**, 3651-3653.
48 C. Hansch, A. Leo and R. W. Taft, *Chem. Rev.*, 1991, **91**, 165–195.
49 J. A. Montgomery, M. J. Frisch, J. W. Ochterski and G. A. Petersson, *J. Chem. Phys.*, 1999, **110**, 2822–2827.
50 C. Dell'Erba, A. Mele, M. Novi, G. Petrillo, F. Sancassan and D. Spinelli, *J. Chem. Soc., Perkin Trans. 2*, 1990, 2055–2058.
51 S. E. Varjosaari, J. P. Hess, P. Suating, J. M. Price, T. M. Gilbert and M. J. Adler, *Tetrahedron Lett.*, 2015, **56**, 642–645.
52 S. Z. Vatsadze, Y. D. Loginova, G. dos Passos Gomes and I. V. Alabugin, *Chem. Eur. J.*, 2017, **23**, 3225–3245.
53 R. Dennington, T. A. Keith and J. M. Millam, *GaussView, Version 5*, Semichem Inc., Shawnee Mission, KS, 2009.
54 M. J. Frisch, G. W. Trucks, H. B. Schlegal, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, G. Scalmani, V. Barone, B. Mennucci, G. A. Petersson, H. Nakatsuji, M. Caricato, X. Li, H. P. Hratchian, A. F. Izmaylov, J. Bloino, G. Zheng, J. L. Sonnenberg, M. Hada, M. Ehara, K. Toyota, R. Fukuda, J. Hasegawa, M. Ishida, T. Nakajima, Y. Honda, O. Kitao, H. Nakai, T. Vreven, J. J. A. Montgomery, J. E. Peralta, F. Ogliaro, M. Bearpark, J. J. Heyd, E. Brothers, K. N. Kudin, V. N. Staroverov, R. Kobayashi, J. Normand, K. Raghavachari, A. Rendell, J. C. Burant, S. S. Iyengar, J. Tomasi, M. Cossi, N. Rega, J. M. Millam, M. Klene, J. E. Knox, J. B. Cross, V. Bakken, C. Adamo, J. Jaramillo, R. Gomperts, R. E. Stratmann, O. Yazyev, A. J. Austin, R. Cammi, C. Pomelli, J. W. Ochterski, R. L. Martin, K. Morokuma, V. G. Zakrzewski, G. A. Voth, P. Salvador, J. J. Dannenberg, S. Dapprich, A. D. Daniels, Ö. Farkas, J. B. Foresman, J. V. Ortiz, J. Cioslowski, D. J. Fox and Gaussian Inc., *Gaussian 09, Revision B.01*, Wallingford CT, 2009.